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Immunocytes of Composite Tissue Allografts Express Elevated Levels of TGFβ mRNA and Protein During Chronic Rejection

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CHRONIC REJECTION after transplantation, including experimental composite tissue grafts, is a major cause of late allograft dysfunction. Recently, we have shown that chronic rejection of rat hind limb transplants results in significant structural and immunological changes.[1] A cytokine which is known to exert a variety of immunoregulatory effects is transforming growth factor beta (TGFβ). This pleiotrophic factor has been shown to be expressed in normal skin.[2] Most of its effects result in immunosuppression, but mitogenic effects are also described. Additionally, it has been implicated in causing hypertrophy of cardiac muscle as well as being involved in the onset of fibrosis.[3] The aim of this study was to investigate whether the expression of TGFβ by infiltrating immunocytes is effected during chronic rejection after composite tissue transplantation and where TGFβ mRNA expressing cells and the TGFβ protein are localized.

MATERIALS AND METHODS

Chronic rejection after total hind limb transplantation (Lewis, RT^{1l}→Brown Norway, RT^{1n}; $n = 5$) was achieved by a limited course of immunosuppression (FK 506, 1 mg/kg/d) for 100 days. Syngeneic animals (Lewis→Lewis; $n = 5$) served as a control. Following 30 days of discontinuation of the immunosuppression, skin and muscle biopsies were taken. The local expression of TGFβ mRNA was investigated using in situ hybridization with S^{35} radiolabelled cDNA probes on 5 μm skin and muscle sections of syngeneic and chronically rejecting transplants snap-frozen on postoperative day 130. Immunohistochemistry was used for localization of the TGFβ protein in those sections.

RESULTS

Upon in situ hybridization, a strong upregulation of TGFβ mRNA expressing cells was detected in sections of chronically rejecting allografts in both skin and muscle compared to the syngeneic controls. TGFβ-positive cells were localized to specific infiltrating immunocytes within the skin and especially the muscle. Corresponding to these findings, the TGFβ protein was detected in the same areas, with a stronger signal compared to the syngeneic controls. TGFβ protein was found, furthermore, in the smooth muscle layer and endothelial cells of blood vessels. The muscle fibers showed no signal for TGFβ. Biopsies from the contralateral leg as well as from syngeneic animals showed no signal or only a weak signal.

CONCLUSION

TGFβ expression is upregulated during chronic rejection after hind limb transplantation in both skin and muscle. Immunoregulation and fibrosis are two of a variety of effects that can be caused by this pleiotrophic protein. These findings suggest a potential role for TGFβ in the immune response during chronic rejection of composite tissue allografts, in which TGFβ may counteract this process since most of the in vivo effects result in immunosuppression. Additionally, TGFβ could induce fibrosis, and thus influence the structural and functional changes that are observed during the rejection process. The modulation of TGFβ might be useful for the improvement of graft survival during chronic rejection.

REFERENCES

1. Llull R, Murase N, Ye Q, et al: Transplant Proc 27:164, 1995
2. Fealy MJ, Most D, Huie P, et al: Transplantation 59:1475, 1995
3. Waltenberger J, Wanders GJ, Fellstrom B, et al: J Immunol 151:1147, 1993

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